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Autologous Hematopoietic Stem Cell Transplantation as a Treatment Option for Aggressive Multiple Sclerosis

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Abbreviations *aHSCT* autologous hematopoietic stem cell transplantation · *CIS* clinically isolated syndrome · *CNS* central nervous system · *Cy* cyclophosphamide · *EAE* experimental autoimmune encephalomyelitis · *EBMT* european group for blood and marrow transplantation · *EBV* Epstein-Barr virus · *GA* glatiramer acetate · *INF-β* interferon- β · *MS* multiple sclerosis · *OCB* oligoclonal bands · *SPMS* secondary progressive multiple sclerosis · *PML* progressive multifocal leukencephalopathy · *PPMS* primary progressive multiple sclerosis · *RRMS* relapsing remitting multiple sclerosis · *TRM* transplant-related mortality

Opinion statement

Despite the development of several injectable or oral treatments for relapsing-remitting multiple sclerosis (RRMS), it remains difficult to treat patients with aggressive disease, and many of these continue to develop severe disability. During the last two decades autologous hematopoietic stem cell transplantation (aHSCT) has been explored with the goal to eliminate an aberrant immune system and then re-install a healthy and tolerant one from hematopoietic precursor cells that had been harvested from the patient prior to chemotherapy. Clinical studies have shown that aHSCT is able to completely halt disease activity in the majority of patients with aggressive RRMS. Research on the mechanisms of action supports that aHSCT indeed leads to renewal of a healthy immune system. Below we will summarize important aspects of aHSCT and mention the currently best-examined regimen.

Introduction

Multiple sclerosis (MS) is considered a prototypical T cell-mediated autoimmune disease that develops based on a complex genetic background and additional environmental triggers such as infection with Epstein Barr virus (EBV), low vitamin D3 levels, and smoking [1, 2]. Pathogenetically, MS is characterized by inflammatory lesions in the central nervous system (CNS), which leads to demyelination, incomplete remyelination, and also to neuronal/axonal damage and glial proliferation. Clinically, MS causes bouts of neurological deficits, in most instances problems of vision, sensory- and motor deficits, but also ataxia, and compromise of neurocognitive and autonomous functions. Relapses usually resolve after days and weeks during earlier stages of the disease, but remit only incompletely or not at all later. MS is heterogeneous in almost every respect including clinical presentation, disease course, imaging findings, the extent and destructiveness of inflammation, and also of neurodegenerative aspects such as demyelination and loss of axons or CNS tissue in general, and finally the response to treatment.

The development of treatments for MS has been very successful, eight drugs are currently available and multiple others either already filed for approval or in late stage clinical development. However, due to the chronic nature of MS, all of these need to be given for long times or forever. Further, all of these have side effects and some of them very serious ones, and, depending on their route of administration, some treatments compromise quality of life. Also, most of these are very expensive, and lead to substantial socioeconomic burden. All currently available disease-modifying treatments of MS are only effective during the earlier stages of disease, when autoimmune inflammation drives the disease process and before too much CNS tissue is irreversibly damaged. The current goals of treatment are therefore to institute MS treatment as early as possible and ideally to halt the disease process completely or at least for as long as possible. In a relatively small percentage of patients MS is so active and relapses or CNS lesions occur so frequently that one has to escalate treatment intensity and start treatments that are more effective, but also have more side effects. Despite these measures relapses can sometimes not be halted, and it is in these patients where we need additional options.

Autologous hematopoietic stem cell transplantation (aHSCT) has evolved as one if not the best option to treat aggressive forms of MS, although acceptance among neurologists is still low. The reasons for this include the fact that mostly secondary- or primary progressive MS patients have been treated in earlier studies, and these responded only incompletely and/or were more prone to develop complications [3]. Further, the prevailing perception is that risk is still very high, ie, mortality ranges up to 10 %, however in recent years, aHSCT has advanced substantially, and transplant-related mortality has been between 1 %–1.5 % since 2000 with the BEAM-ATG regimen [4]. Through tight collaboration between hematologists/transplant specialists and neurologists aHSCT is now more standardized, mortality is within the range of the most active approved therapy, ie, mitoxantrone, the efficacy is likely superior to all other available treatments, its mechanisms are better understood, and aHSCT is probably the only treatment of MS that has to be applied only once with no further need for therapy in the majority of patients, provided that they have been selected carefully. Despite all this, formal proof through a phase III clinical trial is still lacking mainly because testing such a treatment regimen is not supported by pharmaceutical industry, but pursued by academic investigators. Below, we will summarize the most important aspects of aHSCT and give a recommendation, how it should be applied in patients with aggressive disease, which is based on consensus meetings and continuous discussions between investigators in Europe and North America.

MS treatment landscape

Current treatments of multiple sclerosis aim at reducing relapses and preventing or slowing progression of neurological disability in the most common form of MS, which at the time of first disease manifestation such as for example optic neuritis or myelitis is called clinically isolated syndrome (CIS). Once a second relapse occurs that affects a different functional system of the central nervous system

(CNS) or when imaging findings indicate dissemination in time and space, the diagnosis of relapsing-remitting MS (RRMS) is made, which evolves into secondary progressive MS (SPMS) with or without relapses after various time intervals. CIS-RRMS-SPMS affects 80 %–85 % of patients, and only approximately 10 % of cases show disease progression from the beginning (primary progressive MS; PPMS). With respect to disease severity MS runs a benign course and never leads to substantial disability in a minority of patients, and, also relatively rarely MS can be aggressive and lead to death in a few years. MS relapses are treated with high dose intravenous or oral corticosteroids or, in the case of incomplete responses and severe relapses, by plasmapheresis. Available disease modifying drugs are either immunomodulatory or immunosuppressive and hence are effective during the inflammatory, relapsing-remitting phase of the disease.

Different from other neurological diseases such as stroke or Alzheimer's disease, drug development for MS has been very successful, and we have now several first-line treatments for RRMS. Interferon-β (IFN-β) and glatiramer-acetate (GA) are already approved for up to two decades and have moderate efficacy and a favorable, well-known and benign side effect profile. Depending on the country, a recently introduced oral compound, fingolimod, is available as first-line or second-line and orally administered drug. Fingolimod, a sphingosin-1 phosphate receptor agonist, is considerably more effective than IFN-β and GA and overall also well tolerated. Other oral medications are either already approved and will be available shortly (teriflunomide) or have been filed for approval after successful phase III testing (dimethyl-fumarate; laquinimod). The most effective currently available treatment is natalizumab, a humanized monoclonal antibody against CD49d/very late antigen-4 (VLA-4), which is generally also well tolerated, but has led to progressive multifocal leukoencephalopathy (PML), an opportunistic and often fatal infection of the brain by the polyoma virus JC, in over 300 patients so far. A chemotherapeutic agent, mitoxantrone, is used as second-line therapy for more severe RRMS and also for SPMS [5]. Mitoxantrone can only be given for a very limited period of time due to its cardiac toxicity, and secondary leukemias have occurred in up to 2.8 % of MS patients treated with mitoxantrone. Regarding injectable or infusible compounds, several monoclonal antibodies have finished or are about to finish phase III clinical testing (anti-CD25, daclizumab; anti-CD20, ocrelizumab) or have already been filed for approval (anti-CD52, alemtuzumab). Furthermore, various other drugs and treatment approaches including oral- and injectable compounds, tolerization strategies with peptides, peptide-coupled cells, or inactivated autoreactive T cells, immunomodulation with mesenchymal stem cells, and immune reconstitution with autologous hematopoietic stem cells (aHSCT) are currently being tested at different stages of clinical development.

Based on longer time follow-up data from testing IFN-β in CIS, which demonstrated that the conversion to RRMS can be prolonged and that disability evolution can also be slowed by early treatment [6, 7], the prevailing tendency today is to treat patients early. Another strong argument for early treatment is that CNS tissue, despite its ability of functional compensation of certain deficits, has only a very limited capacity for repair at the structural level. As a consequence, MS treatments are most effective during the inflammatory RRMS phase of the disease and have little or no influence once a certain disability level and CNS damage have occurred and when SPMS has begun. It is generally accepted that immunomodulatory or even immuno-

suppressive treatments are not effective in SPMS, and that it is imperative therefore to initiate treatment as early as possible in order to prevent disability accrual as much as possible. However, depending on the drug label and approval of the various therapies in different countries, it may not be possible to start treatment with the most effective drug/s. Furthermore, since MS primarily affects young adults and is a chronic, life-long disease, the route of administration and adverse event profile of the respective treatment, the patient's wish to have children and other aspects have to be taken into consideration. Some of the above drugs, eg, mitoxantrone or alemtuzumab, have long-lasting effects on the hematopoietic system or carry the risk of secondary malignancies and cardiac damage (mitoxantrone) or of secondary autoimmune diseases (alemtuzumab). Natalizumab may lead to PML with increasing risk following 2 years of treatment or even higher risk following prior treatment with chemotherapeutic agents such as mitoxantrone. Hence, treatment decisions, ie, how to begin and escalate therapy and which drug can be given or not after which prior medications, are not easy and need to take into account the severity and prognosis of MS in the individual patient, the prior treatment history, and the long-term adverse profile of the drugs and treatments.

Autologous hematopoietic stem cell transplantation—current status

As reviewed by Saccardi and Mancardi [4], numerous aspects of aHSCT including different stem cell mobilization and conditioning regimens, which patients are best suited, the percentage of progression-free survivors, and which factors contribute to risk have been examined in detail. More than 500 MS patients have received aHSCT in Europe alone in the last 20 years, and follow-up for substantial fraction of these is longer than 10 years. A joint study of the European and American Bone Marrow Transplantation Societies on long-term outcomes after HSCT in MS is currently ongoing. Problems with these data derive from the heterogeneity of the treated patients with respect to disease course and stage, from the fact that most studies have been open, uncontrolled trials, and from differences in the transplant regimens that have been used. Nevertheless, the most important aspects, ie, patient eligibility, transplant regimen, and also the mechanistic understanding have advanced substantially, and a consensus has been reached between European and North American investigators that aHSCT is best applied in patients of 45 years or younger, within the first 5 years after diagnosis, with aggressive RRMS, and using the BEAM-ATG regimen. Below, we will provide more detail regarding the most important aspects of patient selection and transplant regimen.

Patient eligibility criteria—who is a candidate?

From the above reasons treatment responses to aHSCT have been less clear in SPMS patients, and the fraction of patients, who show sustained improvement for more than one EDSS point is much larger in RRMS patients [8]. There is currently no doubt that aHSCT is most effective in highly active RRMS patients. Identification of MS patients, who are

best suited for aHSCT, is one of the most important aspects for successful treatment. Patients should still be in the early stages of the disease, ie, ideally within 5 years after diagnosis, and have a high risk for rapid disease progression, ie, have aggressive MS [9]. Reliable prognostic predictors for an aggressive course are, however, still lacking, and so far the best approach is to combine clinical relapse activity, MRI activity and failure to respond to first-line- and/or second line treatment. In a recent manuscript that summarized the consensus among specialists, who pursue aHSCT, highly active RRMS has been defined as follows:

- at least one severe clinical relapse (ΔEDSS$\geq$1 with Functional Severity Score (FSS) $\geq$2 in motor, cerebellar, or brain stem function in the year prior to evaluation,

- $\geq$1 gadolinium-positive (Gd+) lesion of diameter $\geq$3 mm or accumulation of $\geq$0.3 T2 lesions/month in 2 consecutive MRI 6–12 months apart.

Furthermore patients qualify for aHSCT after failure of conventional best treatment, which is currently natalizumab. Whether aHSCT can already be considered after failure of first line therapy and prior to further escalation steps is still under discussion [9], but unlikely to be accepted by a broader number of MS neurologists before formal proof of the superiority of aHSCT over most active conventional therapies has been provided.

Patients considered for aHSCT should be younger than 45 years. This is based on increasing transplant-related mortality in patients aged over 40 years [3], which may be related to immunosenescence and/or less efficient immune reconstitution in MS patients above this age [3]. Furthermore, patients over 40 years are at increased risk to shift quicker to the secondary progressive phase of the disease [10], during which the benefit of aHSCT is clearly reduced. The path of treatment escalation and eligibility criteria are briefly summarized in Fig. 1.

Stem cell mobilization and conditioning regimen

The main assumption of how aHSCT acts is that an aberrant immune system that underlies the autoimmune disease is eliminated by chemotherapeutic drugs and subsequently a new and again tolerant immune system is installed by infusing the patient with his/her own hematopoietic stem cells that have been collected prior to conditioning. Along this rationale one important question is how intense and complete the conditioning regimen has to be, ie, if most of hematopoietic stem cells need to be eliminated (myeloablative) or if it is sufficient to deplete lymphocytes (lymphoablative). The former absolutely requires stem cell rescue and carries substantially higher risk. Several transplant regimens including more intense ones with whole body irradiation, with different chemotherapeutic agents, changes in graft mobilization and/or manipulation, or also less intensive lymphoablative regimens have been explored during the last 20 years (for review, see reference [4]). Careful assessment and continuous discussion among aHSCT specialists have led to consensus that the

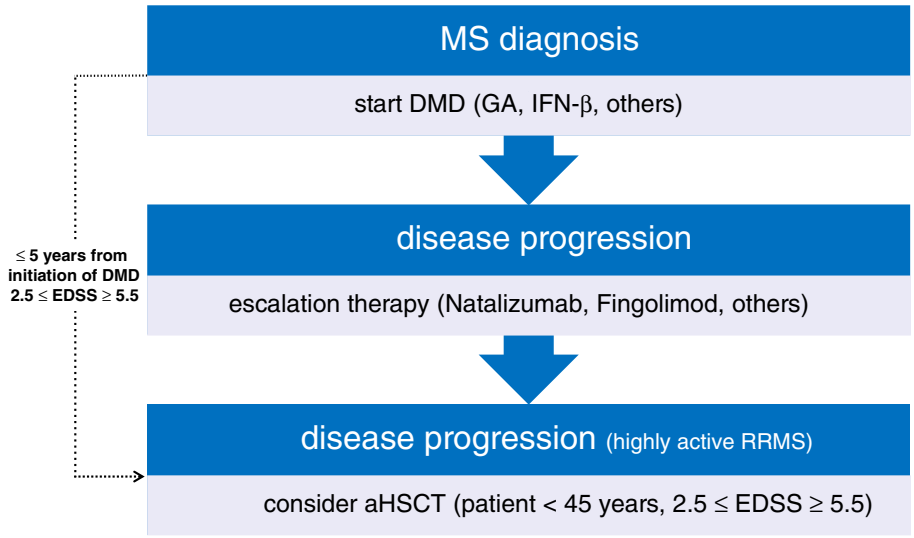

Fig. 1. Proposed therapy algorithm in multiple sclerosis (Adapted from Mancardi et al. [8]).

best experience currently exists for the BEAM-ATG regimen, which has been used most frequently in Europe in the last 15 years. Peripheral Blood Haematopoietic Stem Cells (PBSC) are collected by leukapheresis after mobilization by cyclophosphamide (2-4 g/m2 total dose over 1–2 days) and subsequent treatment with granulocyte colony-stimulating factor (G-CSF, 5–12 µg/kg/day) [4, 11]. Stem cell mobilization is also possible with G-CSF alone, but may lead to MS relapses [12]. Including cyclosphosphamide not only increases the efficacy of mobilization, but probably also reduces the number of autoreactive T cells and inflammatory activity prior to harvesting stem cells. HSCs are then collected and cryopreserved until transplantation. Experience has shown that a number of 3×10^6 $CD34^+$ cells /kg are safe for the patient [4]. The intermediate-intensity conditioning regimen BEAM consists of 300 mg/m^2 carmustine, at day -6200 mg/m^2 etoposide and 200 mg/m^2 cytarabine at days -5 to day -2 and 140 mg/m^2 melphalan at day -1. This regimen has shown a good safety/efficacy profile in lymphoproliferative diseases and in MS was associated to peri-transplant in vivo iv anti-thymocyte globulin (ATG, 5–7.5 mg/kg in 2–3 days) (Fig. 2) [3]. Some studies tested a lymphoablative low-intensity regimen (cyclophosphamide and ATG/alemtuzumab), which shows reduced toxicity, but is less effective in stopping relapse- or in general inflammatory CNS activity (MRI) [13]. To reduce autoreactive lymphocytes from the graft different methods have been used, including ex-vivo positive selection of $CD34^+$ cells prior to cryopreservation. In MS, an evidence of a clinical benefit of graft manipulation is lacking and mobilization with cyclophosphamide plus ATG administration as in-vivo T-cell depletion is now the preferred standard [9]. Besides BEAM-ATG, for which the experience is currently most extensive, a regimen with busulphan/cyclophosphamide/ATG combined with positive selection of $CD34^+$ cells of the graft emerges as another highly effective aHSCT protocol with a good safety profile after the shift to low-dose, iv busulphan [14].

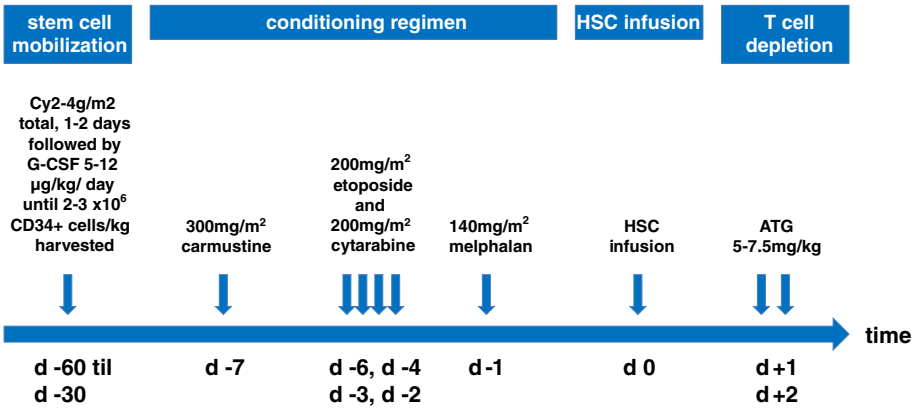

Fig. 2. Protocol for BEAM/ATG regimen in aHSCT. Stem cell mobilization with cyclophosphamide (Cy) and G-CSF. Conditioning regimen with carmustine, etoposide, cytarabine, and melphalan, followed by autologous stem cell infusion. In vivo T cell depletion is performed by anti-thymocyte globulin (ATG).

Adverse events and risks of aHSCT

The transplant-related mortality (TRM) is the major risk for patients undergoing aHSCT in autoimmune diseases (AD). During the first years (1995–2000) TRM was around 7.4 % but decreased from 2001–2007 to 1.3 % [4] as experience of the centers have increased, patient selection improved and toxicity of the protocols decreased. Higher toxicity was observed with regimens including oral busulphan or whole body irradiation [3]. Conversely low-intensity treatments posed less toxicity, but were also less effective for preventing relapses of MS [15]. Pro and contra of myeloablative vs nonmyeloablative regimens are still controversially discussed [9, 16].

Early side effects are mostly caused by immunosuppression and include neutropenic fever, sepsis, CMV reactivation, urinary tract infection, gastroenteritis, pneumonia, and generalized HSV infection as expected for aHSCT in other diseases [3]. Other adverse events were allergy to ATG, engraftment syndrome, veno-occlusive disease, thrombotic thrombocytopenic purpura and mucositis, especially in the late phase [3]. Typical long-term risks are development of a secondary autoimmune disease (about 12 % of patients underwent aHSCT for autoimmune disease develop a secondary autoimmune disease after a median of 22 months [17]). Only one case with a secondary malignancy out of 345 MS patients registered in the EBMT database from 1997–2007 was described [18].

Immune reconstitution and mechanisms of aHSCT

The mechanisms of action of aHSCT are still only partially understood. The clinical experience of long lasting and complete cessation of disease activity with improvement of clinical disability in many patients after aHSCT during the early stages of RRMS suggest that the above premise, ie, elimination of an

aberrant immune system and de novo development of a tolerant/normal immune system, is met and that aHSCT comes close to even curing the disease [4, 18, 19]. Data from Muraro et al demonstrated in a prospective study of 7 patients receiving an intense conditioning regimen (total body irradiation and cyclophosphamide), support this notion and showed that the newly emerging T cell repertoire comes from recent thymic emigrants and is not only broader, but also indicated complete renewal [20]. Numbers of B cells, NKTs, and CD3+ T cells were normal with inverted CD4/CD8 ratio within the first 3 months after aHSCT, whereupon the extra-thymic pathway seems to be predominant in the first year after aHSCT [20–23]. At this stage lymphopenia causes homeostatic proliferation, whereas myelin-reactive T cells undergo activation-induced cell death [24, 25]. While replenishment of CD4+ T cells lags behind other immune cells during the first year, their renewal picks up during the second year and overall T cell diversity increases [26, 27]. Further, similar numbers of myelin basic protein-reactive T cells were observed after autologous HSCT when compared with before the procedure, but after 12 months these MBP-reactive T cells showed a more heterogeneous epitope recognition pattern than at baseline [28]). Studies in experimental autoimmune encephalomyelitis (EAE), the main animal model of MS, demonstrated an increased frequency of regulatory T cells, a shift in T cell myelin epitope recognition and a reduction of anti-myelin antibodies after syngeneic bone marrow transplantation [29]. While the above data on T cell repertoire renewal and elimination of autoreactive T cell clones are relatively complete, there is overall less data on how completely the B cell repertoire and antibody profiles are exchanged following aHSCT. Since oligoclonal immunoglobulin G bands (OCB) in the cerebrospinal fluid are a diagnostic hallmark of MS and may indicate a certain degree of compartmentalization of the autoimmune process in the CNS, it would be important to examine if they disappear as well as comprehensive profiling of the B cell/antibody repertoires before and after aHSCT. Preliminary data indicate that OCB may disappear after aHSCT and remain in some patients, but these studies are not conclusive at present. When considering the homing of long-lived plasma cells to the bone marrow and the relative resistance of these cells to chemotherapy, it is possible that even intense regimens may not completely eliminate them, and that it will then depend on the question if continued or new disease activity requires T cells or can independently be driven by pathogenic B-/plasma cells alone. That antibody-producing cells survive the conditioning regimen is indicated by the at least partially maintained vaccination status to some antigens after aHSCT (for recommendations regarding vaccination after HSCT see Ljungman et al [30]). Mechanistic reasons for the poor/incomplete response to aHSCT in patients, who have secondary- or primary progressive disease [3], may relate to the extent of prior damage of CNS tissue, which is then predestined to slowly progressive neuronal loss and/or to chronic activation of microglial cells and maturation of reactive astrocytes from glial progenitors in response to the inflammatory injury in the CNS as shown by Cassiani-Ingoni et al. in the EAE model [31]. According to their data microglia is not or only incompletely affected by aHSCT.

An important question that relates to the assumption of immune reconstitution as basis for the efficacy of aHSCT is if it is likely that patients may re-

develop MS despite successful immune repertoire exchange since they receive their own stem cells, which carry in principle the genetic risk factors that predispose to MS. Clinical experience shows that a small subgroup of patients indeed re-develop MS even after receiving the currently best aHSCT regimen [4], however, it is currently not clear if these patients: (a) carry a higher number of MS risk loci than those, who remain free of new activity; (b) if they encountered environmental triggering factors; or (c) if the prior damage of CNS tissue and subsequent death of cells releases autoantigens and this is involved in new disease activity. These questions should be addressed in detail in the future. As a first step towards examining these issues Lutterotti et al studied the genome-wide gene expression and microRNA profile of CD34+ hematopoietic precursor cells of MS patients and healthy donors to address if alterations are already found in these precursor cells. Their results indicate that there are no significant differences between CD34+ hematopoietic progenitor cells of MS patients and healthy donors [32].

Summary and future directions

During the last two decades aHSCT and its various aspects have been explored in detail, and the experience can be summarized as follows. Based on casuistic evidence, data from open and uncontrolled studies and the yet unpublished controlled comparison of BEAM-ATG vs mitoxantrone aHSCT (ASTIMS) leads to complete cessation of disease activity in the vast majority of RRMS patients with aggressive MS fulfilling the above criteria. Furthermore, the ASTIMS data shows even in a small phase IIb study that aHSCT is significantly more effective than one of the most effective, currently available drugs, mitoxantrone (Mancardi G, Saccardi R, personal communication). Furthermore, the risk of transplant-related mortality has dropped to acceptable levels in this population of patients. Mechanistic studies support the rationale of aHSCT to abrogate an autoreactive immune repertoire and re-install a healthy immune system. Despite these clear advances, acceptance of aHSCT in the neurological community remains low probably due to the continuing false perception of unacceptable risk, the complexity of the treatment, which requires close interaction between MS neurologists and hematologist/transplant specialist, and the increasing number of treatments that offer a good compromise of acceptable benefit/risk ratio and easier administration. Despite the latter, existing data indicate that even the most effective available drugs need to be given continuously, that particularly the most effective therapies carry substantial risk, and that they are expected to be inferior to aHSCT regarding their efficacy. Further, aHSCT is a one time treatment with no need for continuing immunomodulation, and, when considering the cost of currently available treatments of up to 45,000 USD/year, will lead to substantial socioeconomic benefit in patients with aggressive MS. What is needed most, however, is definitive evidence for superiority of aHSCT over best available therapy of MS by a controlled, multi-center trial. Following several years of intense discussions among European and North-American investigators the main aspects of such a trial have now been outlined and recently published [9]. Since a phase III trial of aHSCT vs conventional treatment will not be sponsored by industry, the main challenge ahead is

to raise interest and funds from public organizations, eg, the European Community in order to provide the missing evidence.

Conflict of Interest

Nikolai Pfender declares that he has no conflict of interest.
Riccardo Saccardi declares that he has no conflict of interest.
Roland Martin has received support for scientific projects from Biogen Idec and during the last five years recompensation for presentations or advisory function by Teva, Biogen, Merck & Serono, and Genzyme, Sanofi & Aventis.

References and Recommended Reading

Papers of particular interest, published recently, have been highlighted as:
- • Of importance
- •• Of major importance

1. Ebers GC. Environmental factors and multiple sclerosis. Lancet Neurol. 2008;7(3):268–77.
2. Simon KC, Munger KL, Ascherio A. Vitamin D and multiple sclerosis: epidemiology, immunology, and genetics. Curr Opin Neurol. 2012;25(3):246–51.
3. Saccardi R, Kozak T, Bocelli-Tyndall C, Fassas A, Kazis A, Havrdova E, et al. Autologous stem cell transplantation for progressive multiple sclerosis: update of the European Group for Blood and Marrow Transplantation autoimmune diseases working party database. Mult Scler. 2006;12(6):814–23.

4.•• Mancardi G, Saccardi R. Autologous haematopoietic stem-cell transplantation in multiple sclerosis. Lancet Neurol. 2008;7(7):626–36.

Important review article of high interest.

5. Freedman MS, Hughes B, Mikol DD, Bennett R, Cuffel B, Divan V, et al. Efficacy of disease-modifying therapies in relapsing remitting multiple sclerosis: a systematic comparison. Eur Neurol. 2008;60(1): 1–11.
6. Kappos L, Freedman MS, Polman CH, Edan G, Hartung HP, Miller DH, et al. Long-term effect of early treatment with interferon beta-1b after a first clinical event suggestive of multiple sclerosis: 5-year active treatment extension of the phase 3 BENEFIT trial. Lancet Neurol. 2009;8(11):987–97.
7. Comi G, Martinelli V, Rodegher M, Moiola L, Bajenaru O, Carra A, et al. Effect of glatiramer acetate on conversion to clinically definite multiple sclerosis in patients with clinically isolated syndrome (Pre-CISe study): a randomized, double-blind, placebo-controlled trial. Lancet. 2009;374(9700):1503–11.

8.• Mancardi GL, Sormani MP, Di Gioia M, Vuolo L, Gualandi F, Amato MP, et al. Autologous hematopoietic stem cell transplantation with an intermediate intensity conditioning regimen in multiple sclerosis: the Italian multi-center experience. Mult Scler. 2012;18(6):835–42.

Interesting summary of aHSCT experience in Italy.

9.•• Saccardi R, Freedman MS, Sormani MP, Atkins H, Farge D, Griffith LM, et al. A prospective, randomized, controlled trial of autologous hematopoietic stem cell transplantation for aggressive multiple sclerosis: a position paper. Mult Scler. 2012;18 (6):825–34.

Article summarizing the emerging consensus how aHSCT should be applied including proposed design of phase III trial.

10. Scalfari A, Neuhaus A, Daumer M, Ebers GC, Muraro PA. Age and disability accumulation in multiple sclerosis. Neurology. 2011;77(13):1246–52.
11. Pasquini MC, Griffith LM, Arnold DL, Atkins HL, Bowen JD, Chen JT, et al. Hematopoietic stem cell transplantation for multiple sclerosis: collaboration of the CIBMTR and EBMT to facilitate international clinical studies. Biol Blood Marrow Transplant. 2010;16(8):1076–83.
12. Openshaw H, Atkins HL, Chen JT, de Bittencourt PR, Griffith LM, Kerr DA, et al. Multiple sclerosis conference synopsis and discussion: cellular therapy for treatment of autoimmune diseases (October 2005). Mult Scler. 2006;12(6):824–5.

13.• Burt RK, Loh Y, Cohen B, Stefoski D, Balabanov R, Katsamakis G, et al. Autologous non-myeloablative

haemopoietic stem cell transplantation in relapsing-remitting multiple sclerosis: a phase I/II study. Lancet Neurol. 2009;8(3):244–53.

Important study on intermediate intensity regimen demonstrating high efficacy, but also the problem of residual or recurring disease activity.

14. Atkins H, Freedman M. Immune ablation followed by autologous hematopoietic stem cell transplantation for the treatment of poor prognosis multiple sclerosis. Methods Mol Biol. 2009;549:231–46.
15. Atkins HL, Muraro PA, van Laar JM, Pavletic SZ. Autologous hematopoietic stem cell transplantation for autoimmune disease–is it now ready for prime time? Biol Blood Marrow Transplant. 2012;18(1 Suppl):S177–83.
16. Burt RK, Balabanov R, Voltarelli J, Barreira A, Burman J. Autologous hematopoietic stem cell transplantation for multiple sclerosis—if confused or hesitant, remember: 'treat with standard immune suppressive drugs and if no inflammation, no response'. Mult Scler. 2012;18(6):772–5.
17. Daikeler T, Labopin M, Di Gioia M, Abinun M, Alexander T, Miniati I, et al. Secondary autoimmune diseases occurring after HSCT for an autoimmune disease: a retrospective study of the EBMT Autoimmune Disease Working Party. Blood. 2011;118 (6):1693–8.
18. Farge D, Labopin M, Tyndall A, Fassas A, Mancardi GL, Van Laar J, et al. Autologous hematopoietic stem cell transplantation for autoimmune diseases: an observational study on 12 years' experience from the European Group for Blood and Marrow Transplantation Working Party on Autoimmune Diseases. Haematologica. 2010;95(2):284–92.
19. Hamerschlak N, Rodrigues M, Moraes DA, Oliveira MC, Stracieri AB, Pieroni F, et al. Brazilian experience with two conditioning regimens in patients with multiple sclerosis: BEAM/horse ATG and CY/rabbit ATG. Bone Marrow Transplant. 2010;45(2):239–48.
20.•• Muraro PA, Douek DC, Packer A, Chung K, Guenaga FJ, Cassiani-Ingoni R, et al. Thymic output generates a new and diverse TCR repertoire after autologous stem cell transplantation in multiple sclerosis patients. J Exp Med. 2005;201(5):805–16.

Important study showing that aHSCT leads to immune reconstitution of a tolerant immune system.

21. Carreras E, Saiz A, Marin P, Martinez C, Rovira M, Villamor N, et al. CD34+ selected autologous peripheral blood stem cell transplantation for multiple sclerosis: report of toxicity and treatment results at one year of follow-up in 15 patients. Haematologica. 2003;88(3):306–14.
22. Saiz A, Blanco Y, Carreras E, Berenguer J, Rovira M, Pujol T, et al. Clinical and MRI outcome after autologous hematopoietic stem cell transplantation in MS. Neurology. 2004;62(2):282–4.
23. Burt RK, Traynor AE, Pope R, Schroeder J, Cohen B, Karlin KH, et al. Treatment of autoimmune disease by intense immunosuppressive conditioning and autologous hematopoietic stem cell transplantation. Blood. 1998;92(10):3505–14.
24. Muraro PA, Douek DC. Renewing the T cell repertoire to arrest autoimmune aggression. Trends Immunol. 2006;27(2):61–7.
25. Kieper WC, Troy A, Burghardt JT, Ramsey C, Lee JY, Jiang HQ, et al. Recent immune status determines the source of antigens that drive homeostatic T cell expansion. J Immunol. 2005;174(6):3158–63.
26. Douek DC, Vescio RA, Betts MR, Brenchley JM, Hill BJ, Zhang L, et al. Assessment of thymic output in adults after haematopoietic stem-cell transplantation and prediction of T-cell reconstitution. Lancet. 2000;355(9218):1875–81.
27. Hug A, Korporal M, Schroder I, Haas J, Glatz K, Storch-Hagenlocher B, et al. Thymic export function and T cell homeostasis in patients with relapsing remitting multiple sclerosis. J Immunol. 2003;171 (1):432–7.
28. Sun W, Popat U, Hutton G, Zang YC, Krance R, Carrum G, et al. Characteristics of T-cell receptor repertoire and myelin-reactive T cells reconstituted from autologous haematopoietic stem-cell grafts in multiple sclerosis. Brain. 2004;127(Pt 5):996–1008.
29. Herrmann MM, Gaertner S, Stadelmann C, van den Brandt J, Boscke R, Budach W, et al. Tolerance induction by bone marrow transplantation in a multiple sclerosis model. Blood. 2005;106 (5):1875–83.
30. Ljungman P, Engelhard D, de la Camara R, Einsele H, Locasciulli A, Martino R, et al. Vaccination of stem cell transplant recipients: recommendations of the Infectious Diseases Working Party of the EBMT. Bone Marrow Transplant. 2005;35(8):737–46.
31. Cassiani-Ingoni R, Muraro PA, Magnus T, Reichert-Scrivner S, Schmidt J, Huh J, et al. Disease progression after bone marrow transplantation in a model of multiple sclerosis is associated with chronic microglial and glial progenitor response. J Neuropathol Exp Neurol. 2007;66(7):637–49.
32. Lutterotti A, Jelcic I, Schulze C, Schippling S, Breiden P, Mazzanti B, et al. No proinflammatory signature in CD34+ hematopoietic progenitor cells in multiple sclerosis patients. Mult Scler. 2012;18 (8):1188–92.